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THE UTILIZATION BY HUMAN SUB-
JECTS OF NITROGEN, CALCIUM,
AND PHOSPHORUS OF THE NAVY
BEAN (*PHASEOLUS VULGARIS*) WITH
AND WITHOUT A SUPPLEMENT
OF CYSTINE

A PART OF A DISSERTATION SUBMITTED TO
THE GRADUATE FACULTY IN CANDIDACY FOR
THE DEGREE OF DOCTOR OF PHILOSOPHY

DEPARTMENT OF HOME ECONOMICS AND
HOUSEHOLD ADMINISTRATION

By
MARTHA SOPHIA PITTMAN

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THE UTILIZATION BY HUMAN SUBJECTS OF THE NITROGEN, CALCIUM, AND PHOSPHORUS OF THE NAVY BEAN (*PHASEOLUS VULGARIS*) WITH AND WITHOUT A SUPPLEMENT OF CYSTINE*

BY MARTHA S. PITTMAN

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PRAUSNITZ (14) reported the results of a digestion experiment on one man who ate daily for 3 days a diet consisting of 500 grams of navy beans and a liter of beer. Since only about 70 per cent of the beans were digested by his subject, Prausnitz concluded that they were not used economically by the body and recommended moderation in employing them in the diet.

A further attempt was made by Snyder (24) to determine the nutritive value of navy beans. He also studied the results of removal of the skins, of cooking the beans with and without the addition of baking soda, and particularly their effect upon the digestibility of other foods. The beans were baked and fed to active young men. For one set of experiments they were eaten with a simple diet of bread and milk, some of the subjects consuming as much as 500 grams of beans daily. The coefficients of digestibility for the protein on this diet appeared to vary greatly with individuals, ranging from 76 to 87 per cent with an average of 80 per cent. With the addition of butter or oleo to the diet the coefficients were higher. If the skins were not removed, 25 per cent less protein was digested and the loss was still greater if the soda were omitted in the cooking process. As a result of these experiments, Snyder also recommended a moderate use of beans in the diet—not to exceed 120 grams of raw beans daily—and stressed the value of proper combinations and preparation.

Wait (25) performed similar experiments using young men as subjects. Besides navy beans, his diets included pork for seasoning, bread, butter, bananas, and sugar. He wished to approximate a mixed diet believing it to be more favorable to digestion. When beans were used in quantities ranging from 375 to 438 grams daily, Wait obtained coefficients of digesti-

* The experimental data in this paper are taken from a dissertation submitted in partial fulfillment of the requirements for the degree of Doctor of Philosophy in the Ogden Graduate School of Science in the University of Chicago, 1930.

bility of 73 to 78 per cent for the protein. With only 150 to 175 grams daily, the coefficients were slightly higher, varying from 78 to 81 per cent. He therefore agreed with the previous workers as to the amounts to be eaten and suggested that beans, as he used them, are less completely digested than other common foods.

The more recent literature on the subject deals largely with animal experiments. McCollum, Simmonds, and Pitz (7) found that when beans were used as the sole source of protein for rats, they appeared to be of low nutritive value resulting in stunted growth and a high mortality rate. These authors therefore recommend that navy beans be supplemented by other proteins of better quality and, because of digestive disturbances, that they be used in moderation.

Johns and Finks (4) were unable to obtain good growth in rats with navy beans until the latter were cooked and cystine was added in the proportion of 2 per cent of the protein by weight. They suggest that heating with water may cause some molecular rearrangement which accounts for the greater digestibility or that it may destroy some toxic quality.

Mitchell (9) obtained with rats at a 10 per cent level of intake, the low biological value of 38.4 per cent for the protein of navy beans after steam cooking. He defines the term "biological value" as the percentage of absorbed nitrogen that is not eliminated in the urine. In similar experiments with other foods at this level, milk proteins averaged 84.7 per cent; potato proteins, 66.7 per cent; oat proteins, 64.9 per cent; and corn proteins, 59.6 per cent.

The possible effect of crude fiber on the excretion of nitrogen, calcium, and phosphorus has been recognized for some time. Since navy beans are relatively high in fiber, this point is to be considered when they play an important part in the diet. Mitchell (10) working with rats which received varying amounts of filter paper in a protein-free diet, found fecal nitrogen increased as much as 42 per cent with this type of roughage. Willard, Whitacre, and Blunt (26) comparing results of feeding human subjects two different diets—one apparently higher in fiber than the other—noted lower coefficients of digestibility for the nitrogen in the high-fiber diet.

In contrast to this view, Sjollem (23) observed that increases in indigestible fiber in the diet of rabbits had little effect on nitrogen output. However, the increased fiber did cause a higher fecal loss of calcium and, to a lesser degree, of phosphorus.

Another factor affecting the usage of calcium and phosphorus may be the amount of carbohydrate in the diet. Bergeim (1) reported, as a result of experiments with rats, that when lactose furnished from 25 to 50 per

cent of the calories the absorption of calcium and phosphorus was improved. This was particularly true of calcium. Such carbohydrates as glucose, maltose, and starch, had little or no effect. Bergeim attributed the superiority of lactose to its tendency to form lactic acid in the alimentary tract which is believed to increase the solubility of calcium phosphate. Earlier work indicated that large quantities of glucose and sucrose taken in concentrated form depressed hydrochloric acid secretion making conditions unfavorable to the absorption of calcium and phosphorus.

EXPERIMENTAL PROCEDURE

Because of the unfavorable reports concerning navy beans for human food, an attempt was made to determine their efficiency as the chief source of nitrogen in a diet providing the essentials for nutrition as recognized today. In doing this the conditions were also unusually favorable for observing the utilization of the calcium and phosphorus. The beans were prepared in two different ways and supplemented with cystine in some cases to determine if the addition of this amino acid, which is considered the chief limiting factor in navy beans, would increase their utilization.

The experiment was divided into two main parts. In the first series of investigations the beans were eaten as a thick purée; in the second, they were baked. These will be designated respectively as the Bean Purée and the Baked Bean Series. Each of these was subdivided as follows:

Without added cystine

Preliminary period 3 days

Collection period 10 days (2 periods of 5 days each)

With added cystine

Preliminary period 3 days

Collection period 10 days (2 periods of 5 days each)

Subjects. Five healthy adult women served as subjects, three being used for each phase of the investigation. All were university students engaged in somewhat similar activities involving more or less laboratory and class room work.

Diet. The diet consisted of navy beans, purified butter fat, sucrose, lactose, grape juice, and lemon juice. The quantities used are shown in Table I. Except for Vitamin D it was doubtless adequate unless the quality of protein should prove to be poor.

The protein was reckoned on the basis of 80 per cent of the requirement set by Sherman of 44.4 grams per 70 kilograms of body weight. This low

level was used to permit differences due to the factors under investigation to be detected readily. The beans were used in sufficient quantity to supply from 90 to 94 per cent of the nitrogen, 80 to 85 per cent of the calcium, and from 84 to 95 per cent of the phosphorus of the various diets. The desired number of calories for the different subjects was secured by varying the amounts of fat and sugars consumed.

TABLE I
AMOUNT OF FOOD EATEN BY THE DIFFERENT SUBJECTS

Food	Subject			Subject		
	A	B	C	C	D	E
	gm.	gm.	gm.	gm.	gm.	gm.
Beans, dry	147.5	119.5	138.0	113.8	163.1	120.9
Butter fat	55.5	38.9	44.4	55.5	99.9	55.5
Sucrose	106.3	106.3	81.3	100.0	112.5	100.0
Lactose	68.5	50.0	50.0	50.0	75.9	50.0
Grape juice	600.0	600.0	600.0	600.0	600.0	600.0
Lemon juice	128.2	128.2	128.2	128.2	128.2	128.2

TABLE II
DIET AS CALCULATED FOR THE VARIOUS SUBJECTS

Subject	Protein	Calcium	Phosphorus	Calories per kg.	Total calories	Excess N. base*
	gm.	gm.	gm.			cc.
Bean purée series						
A	33.2	0.333	0.774	36.0	2358	55.22
B	26.9	0.288	0.642	38.0	2037	50.17
C	31.0	0.318	0.729	37.7	2051	53.50
Baked bean series						
C	25.6	0.279	0.615	42.4	2143	49.14
D	36.7	0.358	0.847	39.6	2863	58.02
E	27.2	0.290	0.648	40.4	2167	50.42

* Without added cystine.

Distilled water was used for all purposes. It was taken *ad libitum* during the Bean Purée Series and the amount consumed varied considerably with the individual and the weather. In the Baked Bean Series each subject took approximately 1200 cc. daily in addition to that obtained in the food. This included a small cup of black coffee for two of the subjects each morning.

In the latter half of each series, 1-cystine, in the proportion of 2 per cent of the weight of the calculated protein, was added to the diet. The allowance for each day was sprinkled over the beans at the time of eating. The amount of salt used (C. P. grade) was not controlled but in no case was it excessive.

The beans were purchased in two lots, one for each series of experiments. The day's supply for each individual was prepared separately on the day before it was to be used. After soaking over night, the beans for purée were salted, and brought to the boiling point over the direct heat, after which they were covered and cooked until soft. This required from 4 to 5 hours in a regulated oven set at 250° F. They were then run through a sieve and all pulp and washings returned as completely as possible, so the loss in preparation was negligible. They were divided into 3 portions, one for each meal. The butter fat was added when they were reheated for serving.

The beans for baking, after soaking, were put into individual baking dishes, one for each meal, and baked with the salt at 350 to 375° F. for 10 hours. The butter fat was added for the last 30 minutes of baking.

The remainder of the diet was combined into a fruit punch. The three meals a day therefore consisted merely of beans and punch.

The amount of roughage in the diet was limited to that present in the beans. The acid-base balance was the same for both periods without added cystine. The excess base was unusually high and, even after cystine was added, the diet was still strongly basic.

The calories remained the same for each subject in both series except for the small increase due to the cystine in the latter half of each period. Because of some loss of weight by the subjects during the Bean Purée Series, which averaged 36 to 38 calories per kilogram, the calories were increased in the Baked Bean Series ranging from 39.6 to 42.4 per kilogram for the various subjects.

Samples. The usual precautions were observed in the collection and handling of samples. Aliquot portions of the foods were saved each day. The beans were cooked, dried at 80° C., ground, and thoroughly mixed. The fruit juices were combined and preserved by canning by the cold-pack method using a water bath. Butter fat, sucrose, and lactose were not sampled as they were considered 100 per cent pure and therefore no analyses were thought necessary.

The urine was preserved with toluol and hydrochloric acid. Each day's collection was kept separate until after nitrogen determinations were made. The feces were mixed with acidified alcohol before drying. A composite was made for each 5-day period.

Methods for Analysis. The pH of the urine was obtained colorimetrically. Total nitrogen was determined by the Kjeldahl-Gunning procedure. Calcium was obtained volumetrically by a modified McCrudden method (8) using Shohl's (20) and Shohl and Pedley's (21) suggestions for controlling pH. Total phosphorus was determined gravimetrically by the Neumann method (12) as modified by Lundell and Hoffman (5) and McCandless and Burton (6). Accuracy of technic was proven by analysis of materials of known composition.

RESULTS AND DISCUSSION

The subjects maintained good health throughout the experiment with the exception that mild indigestion, apparently due to formation of gas, was reported by two subjects in the Bean Purée Series. This difficulty disappeared after the first few days. Since one explanation for such gas is the action of bacteria on fiber, more trouble was expected in the Baked Bean Series in which the fiber was not finely divided. However no trouble occurred, perhaps because of the longer cooking period of the baked beans or the fact that the purée was eaten in hot weather when beans are less appetizing.

It was believed the diet supplied an abundance of calories, yet a slight loss of weight occurred with all subjects. It is possible that with the high-fiber diet used in this experiment less food was absorbed than was expected, making the calories insufficient. However, increased calories in the Baked Bean Series did not decrease the loss in weight, so some other factor was doubtless involved.

The diet was deficient in Vitamin D but experimental work indicates that lack of this vitamin in the diet of normal adults for so brief a time as that covered by this investigation would not affect the results materially.

Urine Findings. The pH of the urine varied greatly with individuals and also in the same individual from day to day even though the diet was practically constant in every respect. The urine seemed to increase in acidity when the weather was warm. The addition of cystine lowered the potential alkalinity of the diet but it did not always result in a more acid urine.

Although some exceptions occurred, the average pH of the urine was more acid than the 6.64 suggested by Hawk and Bergeim (3) for a vegetable diet. The explanation may be, as offered by Pickens and Hetler (13), that although the basic elements are high in grape juice they do not increase the alkalinity of the urine as expected.

Feces Findings. The daily wet weight for the feces ranged from 64.3 to

174.7 grams a day. The average for all periods and subjects was 123.7 grams a day which was equivalent to 25.9 grams dry weight. The average amount of residue, with some exceptions, was not so high as was anticipated because of the fiber in the beans. In accordance with expectations, it was slightly lower in the Bean Purée Series where the beans were broken by running through a sieve.

Nitrogen Findings. In these experiments no corrections were made for endogenous nitrogen. However, the results appearing in Tables III and IV are believed to indicate the efficiency of the navy bean as a source of nitrogen as used in these experiments.

The loss of nitrogen in the feces for both series of experiments ranged from 23 to 43 per cent with an average of approximately 34 per cent. In the Bean Purée Series the excretion of nitrogen in the feces varied from 1.10 to 2.71 grams with an average for all subjects of 1.87 grams. In the Baked Bean Series the excretion ranged from 1.45 to 2.41 grams and averaged 1.93 grams. It is thus seen that the loss was practically the same for both methods of cooking. The large amount of fiber in the beans probably explains such high fecal nitrogen on a diet averaging only 5.2 grams of nitrogen per capita per day.

The average coefficient of digestibility ranged from 57 to 77 per cent for the the various subjects. Both of these extremes were obtained on the Bean Purée diet. The average coefficient of digestibility was 65.9 per cent for all subjects for both periods, so no difference can be attributed to the method of cooking.

In contrast to this, Wait (25) reported an average coefficient of 78 per cent for the nitrogen of white beans used in a mixed diet while Mitchell (9) obtained for rats on a bean diet a value of 76 per cent after correction had been made for metabolic nitrogen in the feces. The lower coefficients obtained in this investigation probably are explained to a large extent by the high crude fiber content of the diet. The fact that the beans were eaten in fairly large quantities, furnishing approximately 20 per cent of the calories consumed, may also have been a factor. It may explain too, a somewhat similar value of 70 per cent obtained by Prausnitz (14) when his subject subsisted entirely upon beans and beer. This harmonizes also with Snyder's higher figures of 80 per cent when some milk was used and of 88 to 90 per cent when a greater variety of foods was eaten.

It appears that the nitrogen from the beans, supplemented by the very small quantities obtained from the remainder of the diet, was inadequate to maintain the body in nitrogen equilibrium for any length of time. The nitrogen intake ranged from 4.23 to 6.73 grams per day or 0.082 to 0.093

TABLE III
NITROGEN FINDINGS
Bean Purée Series

Subject	Expt.	Period	Average daily intake				Average daily output					Balance
			Beans	Other foods	Cystine	Total	Urine	Feces	Total	Urine	Feces	
A	Non-cystine	I	gm. 4.71	gm. 0.41	gm. —	gm. 5.12	gm. 3.81	gm. 1.30	gm. 5.11	per cent 74.6	per cent 25.4	gm. +0.01
		II	4.91	0.40	—	5.31	3.53	2.16	5.69	62.0	38.0	-0.38
		Av.	4.81	0.41	—	5.22	3.67	1.73	5.40	68.3	31.7	-0.19
	Cystine	III	5.06	0.38	0.08	5.52	3.19	1.60	4.79	66.6	33.4	+0.73
		IV	5.10	0.38	0.08	5.56	3.94	1.85	5.79	68.0	32.0	-0.23
		Av.	5.08	0.38	0.08	5.54	3.57	1.73	5.29	67.3	32.7	+0.25
B	Non-cystine	I	3.82	0.41	—	4.23	3.68	1.10	4.78	77.0	23.0	-0.55
		II	3.98	0.40	—	4.38	3.67	1.75	5.42	67.7	32.3	-1.04
		Av.	3.90	0.41	—	4.31	3.68	1.43	5.10	72.4	27.7	-0.80
	Cystine	III	4.10	0.38	0.06	4.54	3.82	1.60	5.42	70.5	29.5	-0.88
		IV	4.13	0.38	0.06	4.57	3.35	2.03	5.38	66.3	33.7	-0.81
		Av.	4.12	0.38	0.06	4.56	3.59	1.82	5.40	68.4	31.6	-0.85
C	Non-cystine	I	4.41	0.41	—	4.82	3.60	2.71	6.31	57.0	43.0	-1.49
		II	4.59	0.40	—	4.99	3.53	2.25	5.78	61.1	38.9	-0.79
		Av.	4.50	0.41	—	4.91	3.57	2.48	6.05	59.1	41.0	-1.14
	Cystine	III	4.73	0.38	0.07	5.18	3.27	2.06	5.33	61.4	38.6	-0.15
		IV	4.77	0.38	0.07	5.22	3.05	2.03	5.08	60.1	39.9	+0.14
		Av.	4.75	0.38	0.07	5.20	3.16	2.05	5.21	60.8	39.3	-0.01

TABLE IV
NITROGEN FINDINGS
Baked Bean Series

Subject	Expt.	Period	Average daily intake				Average daily output					Balance
			Beans	Other foods	Cystine	Total	Urine	Feces	Total	Urine	Feces	
C	Non-cystine	V	gm. 4.23	gm. 0.35	gm. —	gm. 4.58	gm. 3.49	gm. 1.96	gm. 5.45	per cent 64.0	per cent 36.0	gm. −0.87
		VI	4.36	0.35	—	4.71	3.14	2.10	5.24	59.9	40.1	−0.53
		Av.	4.30	0.35	—	4.65	3.32	2.03	5.35	62.0	38.1	−0.70
	Cystine	VII	4.38	0.37	0.06	4.81	3.30	1.92	5.22	63.2	36.8	−0.41
VIII		4.33	0.36	0.06	4.75	3.10	1.93	5.03	61.1	38.9	−0.28	
Av.		4.36	0.37	0.06	4.78	3.20	1.93	5.13	62.2	37.9	−0.35	
D	Non-cystine	V	6.07	0.35	—	6.42	5.14	2.13	7.27	70.7	29.3	−0.85
		VI	6.24	0.35	—	6.59	4.75	2.41	7.16	66.3	33.7	−0.57
		Av.	6.16	0.35	—	6.51	4.95	2.27	7.22	68.5	31.5	−0.71
	Cystine	VII	6.28	0.37	0.08	6.73	4.79	1.85	6.64	72.2	27.8	+0.09
VIII		6.18	0.36	0.08	6.62	4.25	1.67	5.92	71.8	28.2	+0.70	
Av.		6.23	0.37	0.08	6.68	4.52	1.76	6.28	72.0	28.0	+0.40	
E	Non-cystine	V	4.50	0.35	—	4.85	3.30	1.89	5.19	63.5	36.5	−0.34
		VI	4.63	0.35	—	4.98	3.69	1.45	5.14	71.8	28.2	−0.16
		Av.	4.57	0.35	—	4.92	3.50	1.67	5.17	67.7	32.4	−0.25
	Cystine	VII	4.64	0.37	0.06	5.07	3.49	2.32	5.81	60.1	39.9	−0.74
VIII		4.59	0.36	0.06	5.01	3.00	1.54	4.54	66.1	33.9	+0.47	
Av.		4.62	0.37	0.06	5.04	3.25	1.93	5.18	63.1	36.9	−0.14	

grams per kilogram. When furnished by such foods as milk, bread and milk, and meat, this amount of nitrogen was found by Rose and MacLeod (17) to be sufficient to maintain a generous positive nitrogen balance.

The results of addition of cystine were not striking. However, all subjects responded favorably to it in the Baked Bean Series and two of the three subjects in the Bean Purée Series. The range for the nitrogen balance of the subjects on the non-cystine diet averaged -1.14 to -0.19 grams for the different periods, whereas with cystine it ranged from -0.35 to $+0.40$ grams. In two cases the average balance became slightly positive; one subject was in approximate equilibrium, and two were somewhat less negative. In the remaining instance, the difference between the non-cystine and cystine periods was negligible, the balance changing only from -0.80 to -0.85 grams.

In three subjects the improvement took place parallel with an increase of nitrogen in the feces which indicated that in these instances, at least, the higher coefficients were not associated with better nitrogen absorption. This work confirmed that of the earlier investigators to the extent that the nitrogen of the beans, prepared as described in these experiments, apparently was not well utilized by the body.

Similarly, Sherman and Winters (18) and Sherman, Winters, and Phillips (19) were unable to secure nitrogen equilibrium on diets in which corn or oats furnished most of the protein of the diet. These results thus appeared to support the generally prevalent idea that vegetable proteins are less adequate, used alone, than those from animal sources. Snyder's statement (24) that navy beans may be eaten in comfort in amounts of 4 ounces or more (dry weight) a day is also confirmed. Further, the findings in this experiment agreed with those of Mitchell (11) and McCollum (7) that navy beans cannot be regarded as a "meat substitute" when used as the chief source of nitrogen in the diet. However, it appeared probable that a diet consisting largely of navy beans as a source of nitrogen may be made adequate with the addition of a sufficient amount of supplementary protein or cystine.

Calcium Findings. Tables V and VI show that the subjects were practically all in slightly negative calcium balance throughout the experiment and there was little tendency to acquire equilibrium as the diet was continued. Cystine apparently had no favorable effect on calcium usage since all subjects but one showed an increasingly negative average balance after it was added to the diet. The utilization of calcium seemed to be about equal for the two methods of cooking, the Bean Purée Series having a slight advantage.

TABLE V
CALCIUM FINDINGS
Bean Purée Series

Subject	Expt.	Period	Average daily intake			Average daily output					Balance
			Beans	Other foods	Total	Urine	Feces	Total	Urine	Feces	
A	Non-cystine	I	gm. 0.287	gm. 0.060	gm. 0.347	gm. 0.042	gm. 0.286	gm. 0.328	per cent 12.9	per cent 87.1	gm. +0.019
		II	0.314	0.061	0.375	0.053	0.538	0.591	9.0	91.0	-0.216
		Av.	0.301	0.061	0.361	0.048	0.412	0.460	11.0	89.1	-0.099
	Cystine	III	0.304	0.061	0.365	0.086	0.460	0.546	16.0	84.0	-0.181
		IV	0.324	0.058	0.382	0.080	0.468	0.548	14.9	85.1	-0.166
		Av.	0.314	0.060	0.374	0.083	0.464	0.547	15.5	84.6	-0.174
B	Non-cystine	I	0.233	0.060	0.293	0.032	0.226	0.258	10.6	89.4	+0.035
		II	0.254	0.061	0.315	0.041	0.382	0.423	9.6	90.4	-0.108
		Av.	0.244	0.061	0.304	0.037	0.304	0.341	10.1	89.9	-0.037
	Cystine	III	0.246	0.061	0.307	0.056	0.362	0.418	13.4	86.6	-0.111
		IV	0.263	0.058	0.321	0.067	0.442	0.509	13.2	86.8	-0.188
		Av.	0.255	0.060	0.314	0.062	0.402	0.464	13.3	86.7	-0.150
C	Non-cystine	I	0.268	0.060	0.328	0.067	0.317	0.384	18.1	81.9	-0.056
		II	0.293	0.061	0.354	0.085	0.391	0.476	17.8	82.2	-0.122
		Av.	0.281	0.061	0.341	0.076	0.354	0.430	18.0	82.1	-0.089
	Cystine	III	0.284	0.061	0.345	0.127	0.351	0.478	26.5	73.5	-0.133
		IV	0.303	0.058	0.361	0.120	0.334	0.454	26.4	73.6	-0.093
		Av.	0.294	0.060	0.353	0.124	0.343	0.466	26.5	73.6	-0.113

TABLE VI
CALCIUM FINDINGS
Baked Bean Series

Subject	Expt.	Period	Average daily intake			Average daily output					Balance
			Beans	Other foods	Total	Urine	Feces	Total	Urine	Feces	
C	Non-cystine	V	gm. 0.255	gm. 0.066	gm. 0.321	gm. 0.094	gm. 0.350	gm. 0.444	per cent 21.2	per cent 78.8	gm. -0.123
		VI	0.281	0.065	0.346	0.119	0.367	0.486	24.5	75.5	-0.140
		Av.	0.268	0.066	0.334	0.107	0.359	0.465	22.8	77.2	-0.132
	Cystine	VII	0.254	0.072	0.326	0.104	0.344	0.448	23.2	76.8	-0.122
		VIII	0.256	0.065	0.321	0.101	0.380	0.481	21.0	79.0	-0.160
		Av.	0.255	0.069	0.324	0.103	0.362	0.465	22.1	77.9	-0.141
D	Non-cystine	V	0.366	0.066	0.432	0.068	0.441	0.509	13.4	86.6	-0.077
		VI	0.403	0.065	0.468	0.105	0.530	0.635	16.5	83.5	-0.167
		Av.	0.385	0.066	0.450	0.087	0.486	0.573	15.0	85.0	-0.122
	Cystine	VII	0.364	0.072	0.436	0.095	0.491	0.586	16.2	83.8	-0.150
		VIII	0.367	0.065	0.432	0.106	0.405	0.511	20.7	79.3	-0.079
		Av.	0.366	0.069	0.434	0.101	0.448	0.549	18.4	81.6	-0.115
E	Non-cystine	V	0.271	0.066	0.337	0.029	0.419	0.448	6.5	93.5	-0.111
		VI	0.299	0.065	0.364	0.035	0.387	0.422	8.3	91.7	-0.058
		Av.	0.285	0.066	0.351	0.032	0.403	0.435	7.4	92.6	-0.085
	Cystine	VII	0.270	0.072	0.342	0.030	0.537	0.567	5.3	94.7	-0.225
		VIII	0.272	0.065	0.337	0.037	0.433	0.470	7.9	92.1	-0.133
		Av.	0.271	0.069	0.340	0.034	0.485	0.519	6.6	93.4	-0.179

The urinary calcium was low averaging 15.6 per cent of the entire output. Fecal excretion was correspondingly high.

The results for the bean calcium were scarcely so favorable as those obtained by Rose (15), Blatherwick and Long (2), and other workers with calcium from vegetable sources. They resembled those of Rose and MacLeod (17) where almonds supplied from 85 to 86 per cent of the calcium of the diet. These workers were unable to obtain calcium equilibrium on so large a proportion of almonds until the calcium per kilogram of body weight was increased from 8 to 12 milligrams, whereas with other vegetable foods and with a smaller per cent of almond calcium, from 5.6 to 7.0 milligrams of calcium per kilogram of body weight were ample. The diet used in these experiments furnished from 5.5 to 6.5 milligrams of calcium per kilogram of body weight of which 80 to 85 per cent was obtained from the beans.

While it must be granted that the loss of calcium was not great and that the diet provided a scant margin of safety, there seemed to be little tendency to reach a state of equilibrium on the diet. Apparently this amount of calcium, when supplied largely by beans, was insufficient for maintenance. It is probable that the difference was not in the calcium itself but was due to its association with indigestible material.

Phosphorus Findings. The average for each period for each subject as seen in Tables VII and VIII showed a slightly negative phosphorus balance. In four out of six cases the negativity decreased during the cystine periods. When the utilization of the nitrogen improved, with two exceptions, that of the phosphorus improved also. The method of cooking apparently was not an important factor.

In five cases the greater part of the phosphorus was excreted through the feces but the differences were less marked than with calcium. The average loss in the urine was approximately 40 per cent, the remaining 60 per cent being excreted in the feces. In all cases the fecal phosphorus increased during the cystine periods but in only three instances were the differences sufficient to appear significant. Whether this increase of fecal phosphorus meant a poorer digestion of the food, it is impossible to state, but in two out of these three cases there was a parallel decrease in fecal nitrogen indicating better absorption of this element at least.

The average phosphorus intake was low, ranging from an average of 0.551 to 0.769 grams per day or 10.3 to 11.5 milligrams per kilogram of body weight. It is doubtful whether equilibrium could be expected with so small an amount in the food. Certainly the quantities used, when supplied by navy beans, were inadequate to maintain the body in phosphorus equi-

TABLE VII
PHOSPHORUS FINDINGS
Bean Purée Series

Subject	Expt.	Period	Average daily intake			Average daily output				Balance
			Beans	Other foods	Total	Urine	Feces	Total	Urine	Feces
A	Non-cystine	I	gm. 0.554	gm. 0.088	gm. 0.642	gm. 0.245	gm. 0.390	gm. 0.635	per cent 38.6	per cent 61.4
		II	0.583	0.092	0.675	0.336	0.596	0.932	49.2	50.8
		Av.	0.569	0.090	0.659	0.291	0.493	0.784	43.9	56.1
	Cystine	III	0.572	0.088	0.660	0.264	0.438	0.702	37.6	62.4
		IV	0.583	0.091	0.674	0.246	0.521	0.767	32.1	67.9
		Av.	0.578	0.090	0.667	0.255	0.480	0.735	34.9	65.1
B	Non-cystine	I	0.449	0.088	0.537	0.335	0.253	0.588	36.0	64.0
		II	0.473	0.092	0.565	0.311	0.408	0.719	57.0	43.0
		Av.	0.461	0.090	0.551	0.323	0.331	0.654	46.5	53.5
	Cystine	III	0.463	0.088	0.551	0.335	0.402	0.737	45.6	54.5
		IV	0.470	0.091	0.561	0.330	0.492	0.822	42.8	57.2
		Av.	0.467	0.090	0.556	0.333	0.447	0.780	44.2	55.8
C	Non-cystine	I	0.518	0.088	0.606	0.368	0.432	0.800	46.0	54.0
		II	0.546	0.092	0.638	0.320	0.511	0.832	38.5	61.5
		Av.	0.532	0.090	0.622	0.344	0.472	0.816	42.3	57.7
	Cystine	III	0.535	0.088	0.623	0.274	0.486	0.760	36.1	63.9
		IV	0.545	0.091	0.636	0.249	0.447	0.696	35.8	64.2
		Av.	0.540	0.090	0.630	0.262	0.467	0.729	36.0	64.0

TABLE VIII
PHOSPHORUS FINDINGS
Baked Bean Series

Subject	Expt.	Period	Average daily intake			Average daily output					Balance
			Beans	Other foods	Total	Urine	Feces	Total	Urine	Feces	
C	Non-cystine	V	gm. 0.462	gm. 0.079	gm. 0.541	gm. 0.294	gm. 0.380	gm. 0.674	per cent 43.9	per cent 56.1	gm. -0.133
		VI	0.485	0.086	0.571	0.251	0.445	0.696	36.1	63.9	-0.125
		Av.	0.474	0.083	0.556	0.273	0.413	0.685	40.0	60.0	-0.129
	Cystine	VII	0.475	0.090	0.565	0.270	0.404	0.674	40.1	59.9	-0.109
		VIII Av.	0.471 0.473	0.092 0.091	0.563 0.564	0.267 0.269	0.430 0.417	0.697 0.686	38.3 39.2	61.7 60.8	-0.134 -0.122
D	Non-cystine	V	0.663	0.079	0.742	0.373	0.476	0.849	43.9	56.1	-0.107
		VI	0.694	0.086	0.780	0.359	0.586	0.945	38.0	62.0	-0.165
		Av.	0.679	0.083	0.761	0.366	0.531	0.897	40.9	59.1	-0.136
	Cystine	VII	0.680	0.090	0.770	0.309	0.539	0.848	36.4	63.6	-0.078
		VIII Av.	0.676 0.678	0.092 0.091	0.768 0.769	0.324 0.317	0.453 0.496	0.777 0.813	41.7 39.0	58.3 61.0	-0.009 -0.044
E	Non-cystine	V	0.490	0.079	0.569	0.254	0.405	0.659	38.6	61.4	-0.090
		VI	0.515	0.086	0.601	0.235	0.366	0.601	39.1	60.9	±0.000
		Av.	0.503	0.083	0.585	0.245	0.386	0.630	38.8	61.2	-0.045
	Cystine	VII	0.503	0.090	0.593	0.223	0.553	0.776	28.7	71.3	-0.183
		VIII Av.	0.501 0.502	0.092 0.091	0.593 0.593	0.252 0.238	0.428 0.491	0.680 0.728	37.1 32.9	62.9 67.1	-0.087 -0.135

librium although some tendency to improved utilization with continued usage was shown by some subjects. This seemed to be associated with somewhat better retention of nitrogen.

The low urinary excretion of both calcium and phosphorus is to be expected because the diet was low in these elements and high in fiber, both of which are believed to increase excretion of these elements by way of the intestine. However, other factors may also be involved.

It is possible, according to Shohl, Bennett, and Weed (22), that the utilization of both the calcium and phosphorus would have been better had the diet been approximately neutral in reaction. On the other hand, the acids of the fruit juices presumably furnished a favorable medium for absorption of both of these minerals by increasing their solubility.

It also is possible that the large amounts of sugar consumed which, including that of the fruit juices, supplied from 50 to 63 per cent of the total calories, may have affected the absorption of the calcium and phosphorus (Bergeim, 1). Approximately 16 to 20 per cent of the sugar calories was supplied by lactose. The benefit that may have been derived from the latter may have been more or less offset by the depressing effects of the other sugars so it is not believed that this factor could have changed materially the results obtained.

SUMMARY

A study was made of the utilization of the nitrogen, calcium, and phosphorus of the navy bean. Five adult women served as subjects. The beans were cooked in two ways—as bean purée and baked beans. They were eaten with and without a supplement of cystine in the proportion of 2 per cent of the weight of the calculated protein.

The diet—consisting of beans, purified butter fat, sucrose, lactose, grape juice, and lemon juice—was presumably adequate except for some deficiency of Vitamin D.

The following tendencies were observed:

The trend of the nitrogen balance was increasingly negative while the beans were eaten without the cystine supplement.

Addition of cystine in the above proportions, although showing some irregularities, apparently improved nitrogen retention slightly.

The calcium and phosphorus of navy beans in the amounts used were, as a rule, unable to maintain the balance of these elements in the body. The phosphorus was utilized somewhat better than the calcium and showed a slightly favorable response to addition of cystine.

Improved utilization of the phosphorus in most cases appeared to parallel better retention of nitrogen. No such correlation was noted between calcium and nitrogen or calcium and phosphorus.

The methods of cooking used apparently had little effect upon the usage of the elements studied.

Navy beans seemingly can be eaten in comfort by healthy people in quantities at least as large as 4 ounces, dry weight, daily.

This work on human subjects appears to support the findings of others that navy beans are not entirely satisfactory as the chief source of nitrogen in the diet.

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LITERATURE CITED

1. Bergeim, O., Intestinal Chemistry: V. Carbohydrates and Calcium and Phosphorus Absorption. *Jour. Biol. Chem.*, 1926, **70**, 35.
2. Blatherwick, N. R., and Long, M. L., The Utilization of the Calcium and Phosphorus of Vegetables by Man. *Jour. Biol. Chem.*, 1922, **52**, 125.
3. Hawk, P. H., and Bergeim, O., Practical Physiological Chemistry, Ninth Edition, Philadelphia, 1927.
4. Johns, C. O., and Finks, A. J., Studies in Nutrition, III. The Rôle of Cystine in Nutrition as Exemplified by Nutrition Experiments with the Protein of the Navy Bean, *Phaseolus Vulgaris*. *Jour. Biol. Chem.*, 1920, **41**, 379.
5. Lundell, G. E. F., and Hoffman, J. I., Notes on the Determination of Phosphorus. *Jour. Ind. and Eng. Chem.*, 1923, **15**, 44.
6. McCandless, J. M., and Burton, J. Q., Sources of Error in the Determination of Phosphoric Acid by the Molybdate-Magnesia Method. *Jour. Ind. and Eng. Chem.*, 1924, **16**, 1267.
7. McCollum, E. V., Simmonds, N., and Pitz, W., Dietary Deficiencies of the White Bean, *Phaseolus Vulgaris*. *Jour. Biol. Chem.*, 1917, **29**, 521.
8. McCrudden, F. H., Determination of Calcium in the Presence of Magnesium and Phosphates: The Determination of Calcium in Urine. *Jour. Biol. Chem.*, 1911-1912, **10**, 187.
9. Mitchell, H. H., The Biological Value of Proteins at Different Levels of Intake. *Jour. Biol. Chem.*, 1924, **58**, 905.
10. Mitchell, H. H., A Method of Determining the Biological Value of Protein. *Jour. Biol. Chem.*, 1924, **58**, 873.
11. Mitchell, H. H., The Protein Values of Foods in Nutrition. *Jour. Home Econ.*, 1927, **19**, 122.
12. Neumann, A., Einfache Veraschungsmethods (Säuregemisch-Veraschung). *Zeitschr. Physiol. Chem.*, 1902, 1903, **37**, 115.
13. Pickens, L. M., and Hetler, R. A., The Effect of Grape Juice on Nitrogen Retention and Urinary Acidity. *Jour. Home Econ.*, 1930, **22**, 44.
14. Prausnitz, W., Die Ausnützung der Bohnen in Darmkanale des Menschen, *Zeitschr. für Biol.*, 1890, **26**, 227.
15. Rose, M. S., and MacLeod, G., Experiments on the Utilization of the Calcium of Carrots by Man. *Jour. Biol. Chem.*, 1920, **41**, 349.
16. Rose, M. S., and MacLeod, G., Experiments on the Utilization of the Calcium of Almonds by Man. *Jour. Biol. Chem.*, 1923, **57**, 305.

17. Rose, M. S., and MacLeod, G., Maintenance Values for Proteins of Milk, Meat, Bread and Milk, and Soy Bean Curd. *Jour. Biol. Chem.*, 1925, **66**, 847.
18. Sherman, H. C., and Winters, J. C., Efficiency of Maize Protein in Adult Human Nutrition. *Jour. Biol. Chem.*, 1918, **35**, 301.
19. Sherman, H. C., Winters, J. C., and Phillips, V., Efficiency of Oat Protein in Adult Human Nutrition. *Jour. Biol. Chem.*, 1919, **39**, 53.
20. Shohl, A. T., The Effect of Hydrogen Ion upon Determination of Calcium. *Jour. Biol. Chem.*, 1922, **50**, 527.
21. Shohl, A. T., and Pedley, F. G., A Rapid and Accurate Method for Calcium in Urine. *Jour. Biol. Chem.*, 1922, **50**, 537.
22. Shohl, A. T., Bennett, H. B., and Weed, K. L., Effect of Varying the Acid-Base Content of the Diet. *Jour. Biol. Chem.*, 1928, **78**, 181.
23. Sjollem, B., Studies in Inorganic Metabolism. II. The Influence of Crude Fiber and of Protein upon Calcium and Phosphorus Metabolism. *Jour. Biol. Chem.*, 1917, **31**, 421.
24. Snyder, H., Human Food Investigations, Univ. of Minn. Agr. Exp. Station Bul., 1902, **74**, 121.
25. Wait, C. E., Studies on the Digestibility and Nutritive Value of Legumes. U. S. D. A. Off. of Exp. Stations Bul. 187, 1907.
26. Whitacre, J., Willard, A., and Blunt, K., Influence of Fiber on Nitrogen Balance and on Fat in the Feces of Human Subjects. *This Journal*, 1929, **2**, 187.

